Anti-herpes Virus Type 1 Activity of Oleanane-Type Triterpenoids

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The anti-herpes simplex virus type 1 (HSV-1) activity of 15 oleanane-type triterpenoids including glycyrrhizin and its sapogenol was examined and their structure–activity relationships were discussed. Although glycyrrhizin which exhibited in vivo efficacy against HSV-1 replication showed moderate in vitro anti-HSV-1 activity, its sapogenol, glycyrrhetic acid, showed 10 times greater action than glycyrrhizin. Therefore, the in vivo anti-HSV-1 activity of glycyrrhizin administered orally could be reasonably attributed to glycyrrhetic acid generated by hydrolysis by intestinal bacteria. Since the activity of soyasapogenol A was less than 1/20 of that of soya-sapogenol B, the hydroxylation at C-21 seemed to reduce anti-HSV-1 activity. Since kudzusapogenol A, abrisasapogenols B and C lacked the activity, the C-29 hydroxy group would eliminate anti-HSV-1 activity. On the other hand, since the methylesters of kudzusapogenol B and glycyrrhetic acid exhibited greater action, a methoxy carboxy group at C-20 might enhance activity.

Key words anti-herpes simplex virus activity; glycyrrhizin; oleanane-type triterpenoid; structure–activity relationship; Leguminosae

RESULTS AND DISCUSSION

The results of anti-HSV-1 activity, cytotoxicity and a selective index are listed in Table 1. Glycyrrhizin (1) which exhibited in vivo efficacy against

MATERIALS AND METHODS

Materials Glycyrrhizin (1) and glycyrrhetic acid (2) were purchased from Tokyo Chemical Ind. Co. (Tokyo, Japan). Compound 3 was prepared in the usual manner. Compounds 4—15 were prepared as described in previous papers. Fetal calf serum (FCS) was purchased from Gibco BRL (Grand Island, NY, U.S.A.). Sulfonated γ-globulin (Venilon) was supplied by the Chemo-Sero Therapeutic Institute.

Virus and Cells HSV-1 strain KOS and Vero cells were provided by the Chemo-Sero-Therapeutic Institute.

Antiviral Assays The antiviral activity of test samples on HSV-1 (KOS) was measured by the plaque reduction assay. The detailed procedure was described in a previous paper.

Cytotoxic Assays The anticellular activity was examined as described previously.

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Some triterpenoidal saponins have been reported to show anti-herpes simplex virus type 1 (HSV-1) activity. In a preceding paper we reported the in vitro anti-HSV-1 activity of some leguminous triterpenoidal saponins.

Very recently, Cinatl et al. reported that a leguminous saponin, glycyrrhizin, inhibited replication of the severe acute respiratory syndrome (SARS)-associated virus. Glycyrrhizin also showed potent increase in the survival rate against murine HSV-1 encephalitis. Further, Kurokawa et al. proved that a triterpene sapogenol (non-saccharide part of saponin) itself exhibited oral therapeutic anti-HSV-1 activity in mice. These reports have prompted us to examine the anti-HSV-1 action of triterpenoidal sapogenols together with glycyrrhizin and its sapogenol (glycyrrhetic acid).

Herein, we describe the anti-HSV-1 activity of 15 oleanane-type triterpenoids and discuss their structure–activity relationships.
HSV-1 replication showed moderate in vitro anti-HSV-1 activity. On the contrary, its sapogenol, glycyrrhetic acid (2), showed greater action than 1. The potency was 10 times higher. Further, the potency of its methylester (3) was about three times greater than that of 2. The IC$_{50}$ value (8.1 $\mu$m) of 3 was equal to that of soyasapogenol B (5.6 $\mu$m) which showed the highest activity in the previous experiment.5)

The activity of soyasapogenol A (4) was less than 1/20 of that of soyasapogenol B. Since 4 was a hydroxy derivative of 3 at C-21, the hydroxylation at C-21 would reduce anti-HSV-1 activity. Similarly, kudzusapogenol A (5), a hydroxy derivative of 4 at C-29, did not show any activity. On the other hand, kudzusapogenol B (6) having an $\alpha$-carboxy group at C-20 exhibited greater action than 4. Further, the potency of its methylester (7) was about 2.5 times greater than that of 6. Although the configurations for the carboxy groups of 3 and 7 were opposite, a methoxy carboxy group at C-20 might enhance activity.

Although the IC$_{50}$ value (11.5 $\mu$m) of abrisapogenol E (9) having a hydroxy group at C-30 was about one-half of that of soyasapogenol B, the hydroxy derivative (abrisapogenol B, 10) of soyasapogenol B at C-29 did not show any activity. Since abrisapogenol C (8) also lacked the activity, the C-29 hydroxy group would eliminate anti-HSV-1 activity. Abrisapogenol D (11) did not show any activity, indicating that not only the hydroxylation at C-21 but also the dehydroxylation at C-24 might reduce anti-HSV-1 activity, since the potency of soyasapogenol B having a hydroxy group at C-24 was about 7 times that of sophoradiol.5)

The activity of kudzusapogenol C (12) was slightly more effective than that of 4. Further, soyasapogenol C (13) having a double bond at C-21 and C-22 instead of the hydroxy groups, showed moderate activity. The potency of oleanolic acid (14) and its hydroxy derivative (15) was almost comparable with that of 13. Hence, the hydroxy groups at C-21 and C-22 seemed to not contribute to anti-HSV-1 activity.

The obtained structure–anti-HSV-1 activity relationships are summarized in Fig. 1.

Usually, when crude drugs comprising saponins are ad-

Fig. 1. Structure Anti-HSV-1 Activity Relationships of Oleanane-Type Triterpenoids

REFERENCES AND NOTES

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